

Supplementary Information accompanying:

How warm is too warm for the life cycle of actinopterygian fishes?

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## Supplementary Note

### A. Short-term thermal tolerance studies

There is a vast body of literature on the thermal tolerance of fish that cannot be easily covered in a limited space. We therefore focus on a part the literature that are most relevant to our study.

There are at least three different ways in the literature to measure thermal tolerance of organisms, namely CT (Critical Thermal), CL (Chronic Lethal), and LT (Lethal Temperature) methods<sup>1,2</sup>. The observed thermal limits depend on the method as well as exposure time and acclimation temperature<sup>1,2</sup>. CT and CL are dynamic methods, where environmental temperature is changed at a constant rate until the subject organism loses equilibrium or dies. This rate is high in CT (e.g., 0.1 to 1.0 °C/min) and low in CL (e.g., 1 °C/day to 3 °C/month)<sup>3</sup>. It is known that CTM (CT Maximum) is higher than CLM (CL Maximum) because organisms can tolerate short-term exposure to heat better than long-term exposure<sup>3</sup>. LT is a static method, where the subject organism is left under a constant temperature for a preset time span and then usually returned to its normal temperature once the set time period is past and its fate (recovery or death) is observed<sup>4</sup>. Depending on exposure time, LT<sub>50</sub> (LT at which 50% of the sample survive) may differ greatly<sup>5-7</sup>, as in the dynamic methods. For example, Bull Trout (*Salvelinus confluentus*) survived 23.5 °C over 7 days but 20.5 °C for 60 days<sup>7</sup>.

The difference between short- and long-term heat exposures is substantial, usually amounting to 4 to 5 °C of average difference in the maximum temperature tolerance<sup>1,5</sup>.

Given the context of this study, we are more interested in LT and CL values with long

exposure time than CT values, although CT values are more abundant in the literature given its relative ease of measurement<sup>1,8-15</sup>. Between LT and CL values, the former is preferable given the constancy of temperature.

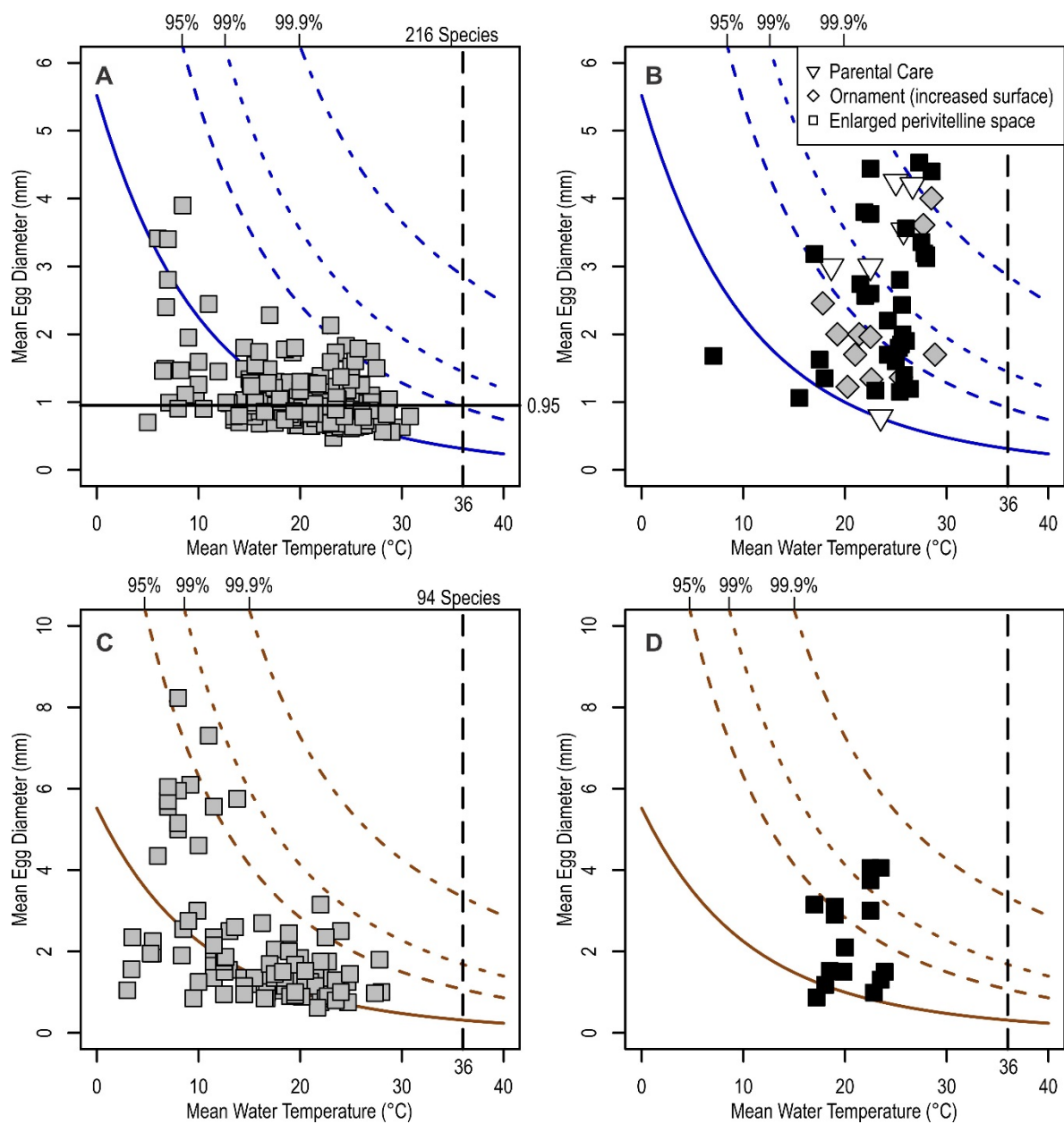
## **B. Thermal tolerance in invertebrates**

It is worthwhile briefly discussing the long-term heat tolerance of marine invertebrates. A study of 36 tropical invertebrates spanning seven phyla found the gastropod *Echinolittorina malaccana* to have the highest long-term heat tolerance, dying after ~5 weeks of exposure to slowly rising temperatures up to 42 °C<sup>16</sup>; other species died before five weeks. Note, however, that these temperatures are for death points, whereas gastropods first lose normal behavior and then fall into heat coma long before thermal death<sup>17</sup>. It was found that loss of normal behavior and heat coma occurred at ~13 °C and ~6 °C lower than thermal death in 11 species of intertidal mollusks from the UK<sup>17</sup>. The highest tolerance of 46.3 °C (~2hr of exposure) was found in *Melarhaphe neritoides*, but it lost normal behavior at 34 °C and fell into heat coma at 38 °C. It is these lower values that seem most relevant to our study. Under natural conditions, these gastropods experience the highest temperatures only when exposed outside water during the day, and they benefit from cooling during the night that allows physiological recovery. Note that hydrothermal vent polychaetes also live at an average temperature of 30 to 35 °C<sup>18</sup>, although they survive short-term exposures (~12 hrs.) of up to 45 °C<sup>19</sup>. Overall, long-term heat tolerance by actinopterygian fishes and invertebrates seem to be similar. Then, paleotemperature data from molluscan fossils probably need to be screen using the same standard proposed for

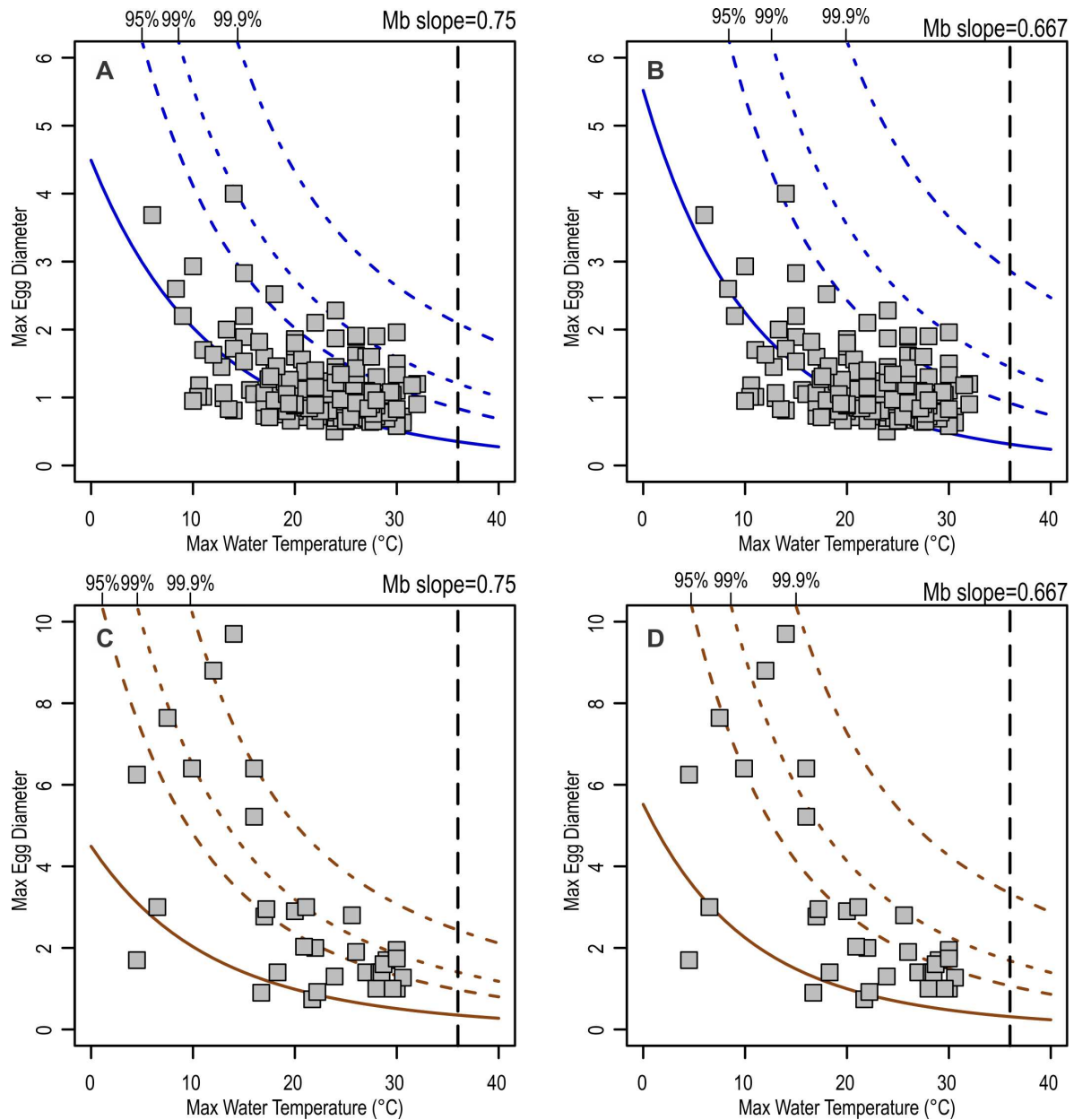
vertebrate fossils in the present paper. However, detailed investigation of thermal tolerance by the broader metazoans is beyond the scope of the present investigation.

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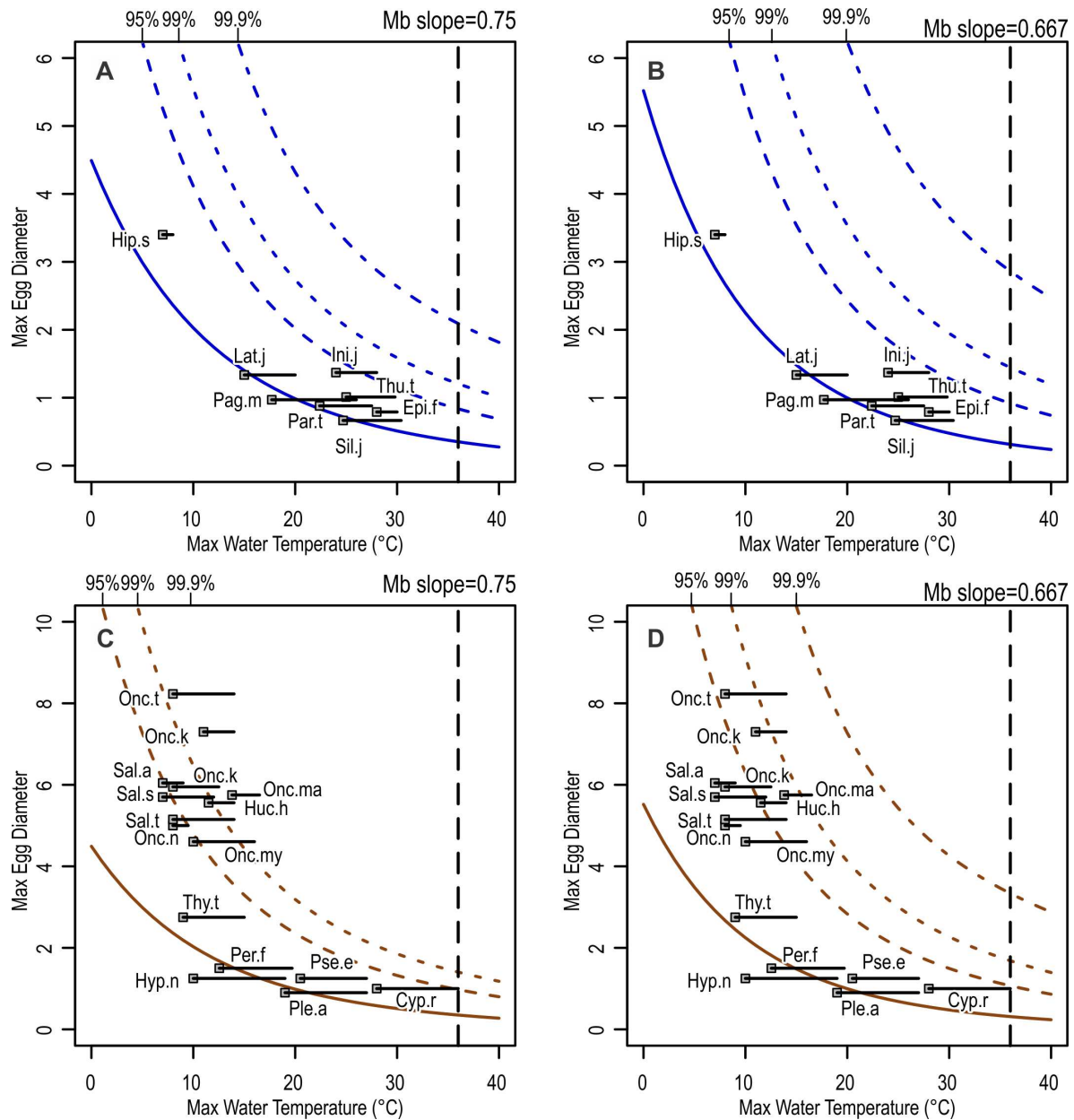
Supplementary Figure S1. Same as Fig. 2 but with a metabolic slope of  $2/3$ . See Fig. 2 caption for details.



Supplementary Figure S2. Same as Fig. 2 and Supplementary Fig. S1 but with maximum values of egg diameter and water temperature for empirical data. The combination of maximum egg diameter and temperature is unrealistic given that eggs laid at the maximum temperature are expected to be minimally small. Therefore, this plot is only given to

facilitate a comparison. A, B, saltwater; C, D, non-salt water. A and C are based on a metabolic slope of  $3/4$ , while B and D assumes a slope of  $2/3$ .





Supplementary Figure S3. Same as Supplementary Fig. S2 but with 50% lethal temperature ranges listed in Supplementary Data. See Supplementary Data for species names.